45. Hydrothermal processing of corn residues: process optimisation and products characterisation

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Abstract
Hydrothermal processing was used as pre-treatment method for the selective solubilisation of hemicellulose from corn residues (leaves and stalks). The raw material was treated at a liquid-to-solid ratio of 10 g/g, under non-isothermal conditions (150-240ºC) and the effect of treatment on the composition of both liquid and solid phases was evaluated. The yields of solid residue and soluble products, e.g., oligosaccharides, monosaccharides, acetic acid and degradation compounds, such as furfural, hydroxymethylfurfural are presented and interpreted using the severity factor (log R_0). The operational conditions leading to the maximum recovery of XOS (53% of initial (arabino)xylan) and for highest glucan content of the solid residue (64%) were established for log R_0 of 3.75 and 4.21, respectively. Under the severest condition 95% of xylan was selectively solubilised and 90% of initial glucan was recovered on the solid residue, making it very attractive for further processing in a biorefinery framework.

Introduction
Corn residues contribute the largest quantities among the agricultural residues in the USA, and are also abundant in China, Brazil and EU. Its chemical composition and low-cost make it an attractive feedstock to be used in a biorefinery framework. Lignocellulosic materials, can be fractionated into their main macromolecular components, cellulose, hemicelluloses and lignin, using environmental friendly processes, originating separated streams, before they can be converted into biofuels and different added-value products [1]. Unfortunately no single technology can accomplish this, and several sequential processes have to be applied. One approach is to initially treat the lignocellulosic materials with a hydrothermal (or autohydrolysis) process which allows a high recovery of soluble hemicelluloses, both oligosaccharides, and to a less extent monosaccharides, while cellulose and lignin can be recovered in the solid phase with minor losses [2-4].

Since autohydrolysis uses water as the only reagent added to the lignocellulosic material, several advantages have been associated with this process as compared to acid pre-hydrolysis, such as low by-product generation, limited problems derived from equipment corrosion owing to the mild pH of reaction media and reduction of operational costs since further neutralisation can be omitted [1,4,5]. The process can be oriented either towards the production of xylo-oligosaccharides (XOS), which are potential ingredients for the pharmaceutical and functional food markets, or to obtain hemicellulose free residues for further valorisation [6-8]. The aim of this work is to study the selective fractionation of corn residues using hydrothermal treatments (autohydrolysis). The effects of pre-treatment on the yield of XOS, monosaccharides and by-products together with the effects on both cellulose and lignin recovery, were evaluated and interpreted using the severity factor (log R_0).

Experimental

Raw material

Corn residues (leaves and stalks) were supplied by Estação Nacional de Melhoramento de Plantas (Elvas, Portugal). The raw material was ground with a knife mill to particles smaller than 6 mm and, after a granulometric characterization, particles smaller than 250 μm were discarded. The material was then homogenised in a define lot, and stored in plastic containers at room temperature.

Hydrothermal processing of corn residues

The hydrothermal treatments (autohydrolysis) were performed in a stainless steel reactor (Parr Instruments Company, USA) with a total volume of 600 ml. Temperature was controlled through a Parr PID controller (model 4842). The raw material was mixed with water in the reactor in order to obtain a liquid-to-solid ratio (LSR) of 10 (kg water/kg dry raw material). The agitation speed was set at 150 rpm and the reactor heated to reach final temperatures ranging between 150ºC and 240ºC (non-isothermal conditions). When the desired temperature was attained, the reactor was rapidly cooled down and the liquid and solid phases were recovered by pressing (up to 190 bar) using a hydraulic press (Sotel, Portugal). The liquid phase was filtered through Whatman #1 filter paper. The solid phase was washed, filtered again, dried at 40ºC and the composition was determined as described below.
The effects of time and temperature on corn residues autohydrolysis were interpreted based on the severity factor, log R_0 [9]:

$$R_0 = \int_0^t \exp\left(\frac{T(t) - 100}{14.75}\right) dt$$

where T the temperature (ºC) is a function of t (time, min), and 14.75 an empirical parameter related with activation energy and temperature.

Analytical Methods

Chemical characterisation of raw material and processed solids

The materials were ground in a knife mill to a particle size smaller than 0.5 mm and the moisture content was determined by oven-drying at 100°C to constant weight. The ash content was determined using NREL/TP-510-42622 protocol [10]. The samples were analysed for glucan, xylan, arabinan and acetyl groups after quantitative acid hydrolysis with 72% (w/w) H_2SO_4 followed by hydrolysis with 4% (w/w) H_2SO_4 [8]. The acid insoluble residue was considered as Klason lignin, after correction for ash. Monosaccharides and acetic acid in the hydrolysates were analysed by HPLC as described below. For extractives determination samples were successively extracted with dichloromethane, ethanol and water with an adapted Soxtec extraction system [11].

Chemical characterisation of liquors and hydrolysates

A sample of the liquors was directly analyzed by HPLC (Waters, USA) for glucose, xylose, arabinose, acetic acid, hydroxymethylfurfural (HMF) and furfural using an Aminex HPX-87H column (Bio-Rad, USA) as previously described [4]. Another sample was hydrolysed with 4% (w/w) H_2SO_4 to convert soluble hemicellulose into their constituent sugar monomers. Oligosaccharides concentrations were calculated from the increase in sugar monomers, as analyzed by HPLC, after acid post-hydrolysis. The term XOS has been used to name the hemicellulose-derived OS made up of xylose units and the possible arabinan-derived oligomers encountered have been included as XOS for the purpose of calculating yields and average compositions.

Results and discussion

Chemical composition of corn residues

Table 1 shows the chemical composition of corn residues (leaves and stalks) used in this work.

Table 1.- *Average macromolecular composition of corn residues.*

Component	% of dry weight
Cellulose (as glucan)	42.1
Hemicellulose	29.2
Xylan	22.9
Arabinan	2.9
Acetyl groups	3.4
Acid insoluble lignin	17.5
Ash	4.2
Extractives	11.6

Cellulose, as estimated from the glucan content, was the major component, whereas hemicellulose, estimated from xylan, arabinan and acetyl groups content accounted for 29.2% of the raw material. This material presented very high polysaccharides content (71.3%) which favourably compares to previously reported values for corn stover [12,13].

Hemicellulose content and specifically the amounts of arabinan, and acetyl groups, were similar to the reported for straws, e.g., wheat and barley [6] and corn stover [12] while the xylan content was slightly higher. Concerning acid insoluble lignin content, the results were in the range of the previously described for less lignified materials such as cereals and grasses. Extractives content was higher than the reported for corn cobs [6,14], but similar to the described for corn stover [15].

Autohydrolysis profile

Figs. 1 and 2 show the variation of xylan, glucan, oligosaccharides, monosaccharides and furan derivatives recovery as a function of the severity factor. As expected, autohydrolysis mainly affected hemicellulosic components (Fig. 1). Xylan hydrolysis becomes important for a severity factor above 2.97, from which it decreased sharply to reach a very significant solubilisation at the severest condition assayed. The highest recovery of XOS (expressed as arabinose substituted xylo-oligosaccharides), was obtained for the severity factor of 3.75 and corresponded to 53.2 g/100 g feedstock (arabino)xylan. In these conditions, 72.1% of the original (arabino)xylan was solubilised and 63.2% was recovered as soluble saccharides (XOS, xylose and arabinose). The XOS yield obtained was lower than the 65% reported for corn cobs autohydrolysis [14,16] but higher then the reported for other straws like wheat and barley straw [6]. Monomeric pentoses yield increased reaching a maximum value of 15.1 g/100 g feedstock (arabino)xylan for log R_0=4.21. For the severest condition there was a sharp decrease in the recovery of those monomeric pentoses, due to degradation reactions. As a consequence furfural concentrations increased to reach up around 20% of initial (arabino)xylan. These results were similar to those reported for wheat straw autohydrolysis [8] up to a severity factor of 3.89, although for higher severities a dramatic increase was observed, which is consistent with the decrease of pentoses.

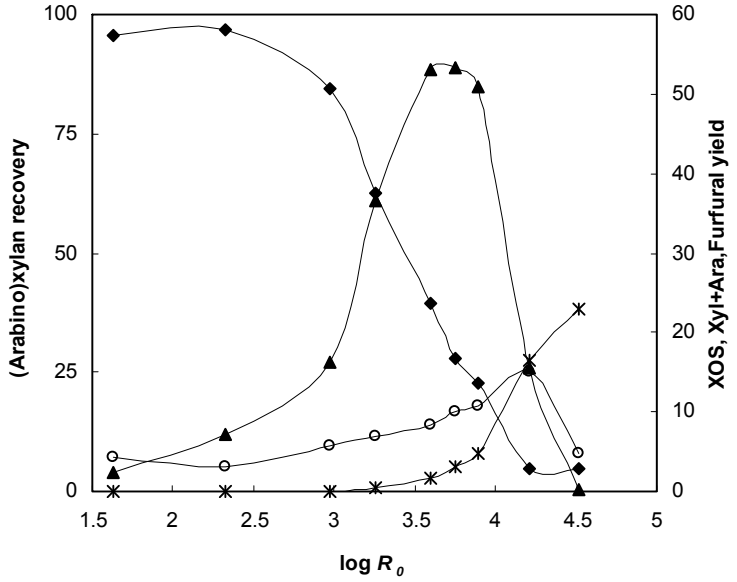

Figure 1. *Recovery of (arabino)xylan and xylo-oligosaccharides, monomeric pentoses and furfural yield as a function of severity after autohydrolysis of corn residues. Results are presented as a percentage of initial (arabino)xylan in the raw material (♦, (arabino)xylan; ▲, XOS; ○, xylose+arabinose; *, furfural)*

In contrast with xylan hydrolysis, glucan essentially remained in the solid phase. However, some oligosaccharides made up of glucose, gluco-oligosaccharides (GlcOS) were recovered reaching a maximum yield of 3.31 g/100 g glucan for severity condition of log R_0=3.60. The maximum yield of glucose was 2.62 g/100 g glucan, and it was achieved at log R_0=2.97 (Fig. 2). In all conditions, HMF concentrations were also low and similar to those reported for the autohydrolysis of other raw materials [4,6,8,17].

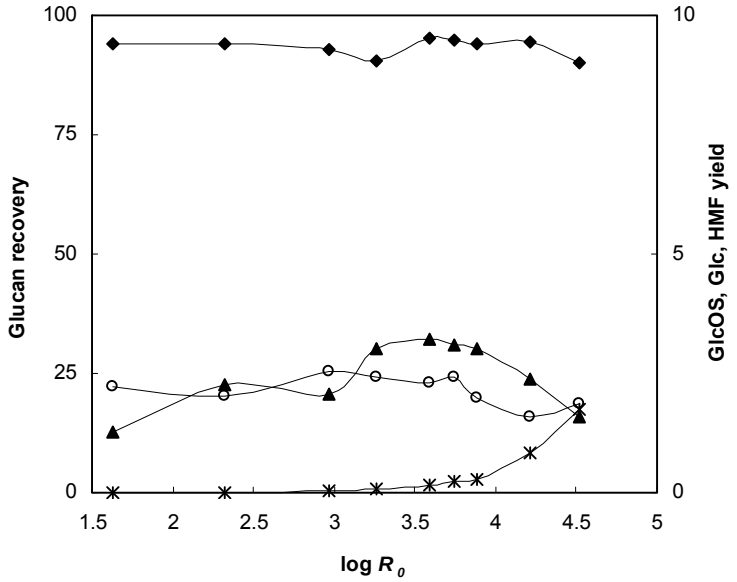

Figure 2. *Recovery of glucan and gluco-oligosaccharides, monomeric glucose and hydroxymethylfurfural yield as a function of severity after autohydrolysis of corn residues. Results are presented as a percentage of initial glucan in the raw material (♦, glucan; ▲, GlcOS; ○, glucose; *, HMF)*

Composition of the liquid phase

The composition of the liquors obtained from autohydrolysis of corn residues under the optimised conditions for XOS production (log R_0=3.75) is presented in Table 2.

Table 2 *- Composition of the liquors obtained from autohydrolysis of corn residues under optimised conditions for XOS recovery (log R_0=3.75).*

Component	Composition (g/l)
XOS	13.30
GlcOS	1.27
AcO	1.25
Xylose	1.88
Arabinose	0.96
Glucose	1.10
Acetic acid	1.30
HMF	0.07
Furfural	0.49

XOS-arabinose substituted xylo-oligosaccharides; GlcOS-gluco-oligosaccharides; AcO-acetyl groups linked to oligosaccharides.

Sugars were recovered mainly in oligomeric form, which is typical for autohydrolysis processes, as the conditions leading to the highest recovery of soluble hemicelluloses do not lead to its complete hydrolysis [18]. Xylo-oligosaccharides are the main oligomeric components of the liquors at this severity condition. Gluco-oligosacchrides and acetyl groups linked to oligosaccharides were also obtained, although in much lower concentrations. All monosaccharides and acetic acid did not exceed 2 g/l, being xylose the main monosaccharide obtained. In these conditions furfural content was low, but it was still the main furan derivative, as HMF appears in almost negligible amounts.

Composition of the solid phase

Table 3 shows the composition of the solid phase obtained for the different severity conditions assayed. For the less severe conditions, the solid solubilisation that occurred was low and the composition of the solid residues remained quite similar after autohydrolysis. However, for log R_0 higher than 2.33 the solid yield decreased sharply to reach about 61.5% for the severest operation condition assayed. This decrease could be mainly correlated to the solubilisation of hemicellulose components. The amount of solubilised xylan increased with severity to reach 95.4%. An almost complete solubilisation also occurred for the other hemicellulose components, showing the efficiency of this treatment towards hemicelluloses.

Glucan was almost not affected by the hydrolytic treatment and a solid residue with a glucan content of 64% was obtained for a severity of log R_0=4.21. The increase of severity only had a minor effect on glucan solubilisation, although the maximum value (9.9%) was obtained for the severest condition. The results obtained suggest that the glucan solubilisation that occurred for the less severe conditions may not be related with cellulose hydrolysis but with other water-soluble components, like non-structural sugars [15]. Low solubilisation of glucan has also been reported for corn cobs autohydrolysis [16,19] and is an advantage for the integral utilization of this raw material in a biorefinery framework.

Table 3. *Effect of severity factor on the solid yield (SY) and polymeric composition of processed solids obtained after autohydrolysis of corn residues.*

	Severity factor, $\log R_0$								
(%)	1.63	2.33	2.97	3.26	3.60	3.75	3.89	4.21	4.51
SY[a]	92.4	91.2	85.0	76.4	69.6	67.8	64.9	62.1	61.5
Xylan[b]	23.0	23.6	22.6	18.9	12.9	9.2	7.7	1.0	1.0
Arabinan[b]	3.7	3.8	3.0	2.2	1.6	1.4	1.4	1.0	1.0
Glucan[b]	42.9	43.5	46.0	49.9	57.6	58.8	61.0	64.0	61.7
Acid insoluble lignin[b]	18.0	17.9	18.9	20.2	21.8	23.6	21.8	27.1	31.0

[a] (g/100 g raw material); [b] (g/100 g processed solids)

Under the present conditions of autohydrolysis no significant removal of lignin was expected to occur. In fact, up to severity factor log R_0=4.21, about 90% of initial acid insoluble lignin was recovered, which favourably compares with the results previously reported for corn cobs [14]. For the most severe condition lignin recovery exceeded 100%. This increase has been associated to the condensation of lignin with sugar and/or sugar degradation products, such as furfural [20,21] to give insoluble reaction products, causing an increase in apparent acid insoluble lignin yield. It is also possible that other compounds such as proteins may contribute to an increase in the acid insoluble lignin (Carvalheiro, unpublish data), although the protein content of corn leaves and stalks is relatively low. This aspect will be subject of further investigations.

Conclusions

Autohydrolysis was highly selective towards hemicellulose enabling a high recovery of XOS, under relatively mild conditions. The optimum conditions were found for 215ºC (log R_0=3.75). Under these conditions, an important glucan enrichment of the solid phase was possible making the processed solids very attractive for further processing.

The chemical composition of corn residues (leaves and stalks) makes it a suitable raw material to be used in a biorefinery framework.

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